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2. Humoral and Cellular Elements in Natural and Acquired Resistance to Typhoid[1,2]

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SIXTY years ago investigators, beginning with Flugge, Nuttall, Buchner, and Ehrlich, had developed a humoral theory of resistance whereby the blood

[1] Paper presented as part of Symposium on "Light from Animal Experimentation on Human Heredity at fifth annual meeting of The American Society of Human Genetics, Ithaca, New York, September 9, 1952.

[2] Journal Paper No. J.-2179 of the Iowa Agricultural Experiment Station, Ames, Iowa. Project No. 1180.

plasma carried immune bodies which precipitated, agglutinated, or otherwise fixed pathogenic elements. Lysins, complements, amboceptors, toxins, and antitoxins were introduced as further concomitants of the humoral elements in the disease reactions. The attitude was one of doubt that phagocytosis of bacteria by leucocytes as observed by Metchnikoff and his group was the determining element in natural resistance. Attempts were made to base a theory of natural immunity on humoral elements known to be products formed in acquired resistance. Research over the intervening years has brought the theories closer together in that as evidence was accumulated it suggested that humoral elements may be the products of the leucocytes. The humoral elements have significance to immunity acquired after contact with the disease. It still remains to be shown if these elements are of importance in the first contact when natural resistance must exercise its effect. Besides the specialized cells and their possible products a cell constituent has become increasingly important in interpreting all life processes. The genes which are carried in the chromosomes of each cell are assuming greater significance to disease resistance. Questions of how these genes act on the organism as a whole and in the disease syndrome are of basic importance to the earlier theories of humoral and cellular elements in resistance. In a few cases a single gene difference is sufficient to make the difference between susceptibility or resistance to a disease. In other cases more genes are involved resulting in a more complex inheritance and resistance graded in its expression. The graded series may arise through differences in either the genotypes of the hosts or the genotypes of the pathogens initiating the disease syndrome.

The level of mortality and morbidity to a constant dose of infectious typhoid in domestic fowl or mice for example, is dependent upon the proper relations between the genetic constitutions of the pathogen and of the host. In a sense, disease severity is built on a square, figure 1. On one side of this square are the genetic constitutions of the hosts. At one end specified doses of the pathogen cause complete mortality to hosts of the highly susceptible constitution. In the middle like doses induce 50 per cent deaths. At the other end, similar doses scarcely affect the activities of the hosts, let alone cause death.

The genetically different typhoid pathogens are on the other side of the square. At one end a genetic line of the organisms is practically innocuous to any strain of the host. In the middle the genetics of the line endow it with capacities to kill most of the susceptible mice, allow the survival of about 50 per cent of the medium susceptible mice, and nearly all of the resistant mice. At the other end the virulence of the pathogen line is so great that some mice of even the resistant strain die when inoculated with the standard dose of organisms.

Over the surface of the square the different genetic constitutions of host and pathogen fit together in such a manner as to give all types of disease

reactions. This key to lock type of mechanism of disease mortality suggests the next steps to be taken if disease is to be understood. Information is needed on what the genes do to the hosts in making them susceptible or resistant.

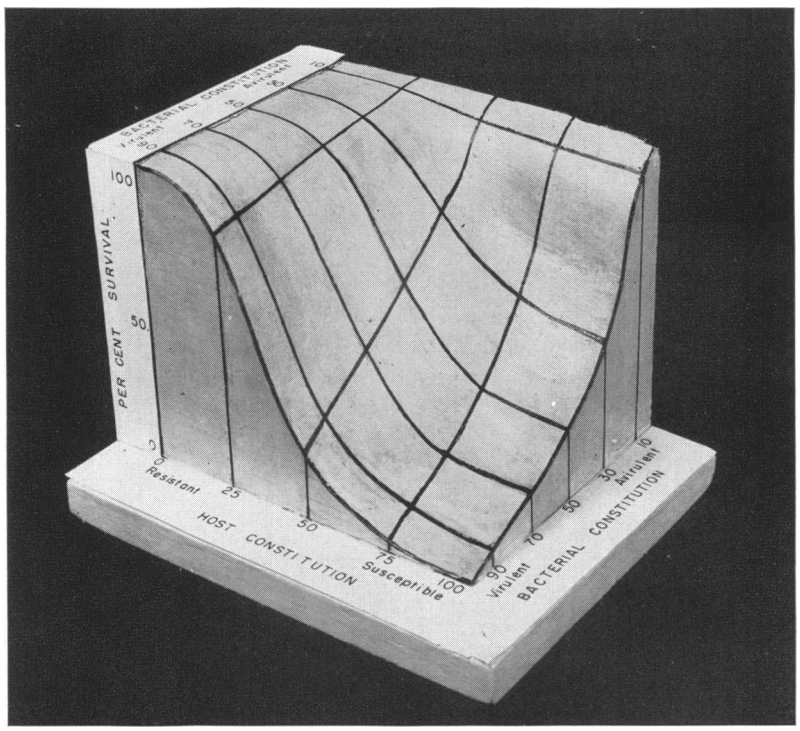


FIG. 1. Model showing interrelation of genotypes of host and pathogen in controlling the severity of a disease. Genotypes of the hosts, ranging from resistant to susceptible, are shown on the front side of the square. The genotypes of the bacteria or other pathogens are shown on the Y axis. The severity of the observed disease produced is shown by the heights above the plane measured in terms of per cent survival of the hosts after the disease has run its course.

Variation in Host Genotypes as a Cause of Variation in Disease Severity

The genetic constitutions of the host cause many different types of disease without any other cause being present. In development from the egg to the infant, in the adolescent, and finally in the senescent adult particular genes may cause morbidity and death to what has up to that time been a well functioning individual. Rh blood factors working in pregnancy, factors for diabetes mellitus operating at any period after birth, the gene for Huntington's chorea coming into expression only after the prime of life has passed are but examples of thousands of genes which have now been studied and shown to be agents of disease in their own right (Gowen 1948).

These genes are capable of extending their effects through a population just as so called infectious diseases spread. At the moment recessive genes

causing dwarf calves and ultimate early death in Hereford, Shorthorn and Angus cattle have become so widespread as to cause real concern. This spread has been facilitated by the heterozygote having a form particularly desired by breeders of feeder and exhibition cattle. Other genes showing like pandemic possibilities as atresia coli in horses and hairlessness in cattle are well known. In man conditions of achondroplastic dwarfs and brachydactyly of the fingers familiar to us today were respectively recorded by the Egyptians around 3500 B.C. and by the Romans during their rise to power. Genes of this type may ultimately be more important for the race than so called infectious pathogens. They are truly cellular elements in which the normal alleles lead to an unrestricted life for the organism and the reciprocal alleles result in morbid conditions of more or less severity.

The purpose of this paper is not, however, to discuss these genes; it is rather to confine ourselves to the interrelations between the genotypes of host and external agents in infection and disease severity. Research on plants and their diseases is on the whole more advanced than that on the animal disease. Two animal investigations should be mentioned as setting a pattern to subsequent studies. In the liver disease of mice caused by the long rod bacillus *B. piliformis*, Tyzzer (1917) observed a case where susceptibility to this disease shows a species difference that is open to analysis since the host species will cross. As the number of mice involved is not stated the case is incomplete but for the *Mus musculus* strain the brown agouti had only two deaths whereas for the *Mus bacterianus* the waltzing strain had 144 deaths. The F_1 to F_4 together with the backcrosses indicated an inheritance of the resistance. Gowen and Schott (1933b) some twenty years later were able to repeat this study, by feeding infected liver material. For *Mus musculus* of 37 mice tested none died; for *Mus bacterianus* of 99 mice tested, 80 died. The F_1 expressed resistance, 14 died in 89 tested. The backcross tests showed segregation according to the parent strain entering the backcross; 4 died to 71 lived, or 5 per cent, when the F_1 was mated to *Mus musculus*; and 38 died to 30 lived, or 56 per cent, when the F_1 was mated to *Mus bacterianus*. For this cross the tentative hypothesis of a single major factor difference between the two strains responsible for the mortality differences expressed by the two species seems reasonable. Other strains of the *Mus bacterianus* may have more complex genotypic differences however. Such differences were shown by a strain derived directly from a stock imported from Peking. The mode of action of the gene complex is not known in either case.

The second case is particularly applicable to this discussion since the inheritance directly affects the humoral element, complement. Rich (1923) in studying Guinea pigs found pigs from which he established a strain free of complement, as distinct from those of normal high complement titer. Genetic analysis showed that the difference was dependent on a single gene pair, the

no complement condition being recessive. The pigs of each group were later infected, spontaneously and by inoculation, with *Pasteurella suiseptica* and *Salmonella cholerae suis* with similar results in each case. Mortality in the complement-free pigs was 77 per cent, while that for the full complement pigs was 20 per cent. Later work showed that the recessive condition altered a hitherto unknown specific portion of the complement thus preventing it from performing its specific function. Here the gene as a cellular element operates through forming a humoral element having a direct part in the disease reaction. But few diseases show such simplicity. Ordinarily many different pathways are important to the full expression of a disease with the consequence that many genes of both host and pathogen are involved.

Typhoid of the Mice

This is true of the typhoid disease of the mouse due to *Salmonella typhimurium*. Within mice there is much variation of genotypes. This variation makes it possible to separate out strains which are resistant, strains of intermediate resistance, and strains which are susceptible to a given line of the pathogen. This separation of the host reaction to typhoid has been accomplished in several ways. In our laboratory, strains are maintained that have the following characteristic survival values when inoculated with 200,000 organisms of our line 11C of *Salmonella typhimurium*; S 86 per cent, RI 82 per cent, Z 58 per cent, K 54 per cent, E 38 per cent, L 12 per cent, and Ba 1.5 per cent. Each survival percentage is based on more than 1000 animals. The typhoid reaction of each strain is as characteristic of the strain as that of any other inherited attribute. Over a period of 15 years, the breeding animals of each successive generation have had no known contact with the disease yet their progeny have maintained their relative resistances in each successive generation.

The genetic effects extend to morbidity as well as mortality. Mice of the resistant *S* strain at the same place in the disease cycle are not as morbid as those of the *Ba* strain, Figure 2.

What Elements are Important to Disease Resistance

In this first contact with the disease, the natural resistance of the strains is not dependent on any demonstrable agglutinins or bactericidal powers present in the blood. In fact agglutinins, i.e., appear in the blood so late in the disease cycle as to be seemingly ineffective as agents to modify the initial course of the disease. The lack of humoral elements of importance to the disease reaction is brought out in other ways.

Matings of males of the most resistant strains with females of the most susceptible strains and vice versa were shown by Hetzer (1937) to give progeny with similar resistance; resistant male x susceptible female 87 per cent survive

for 141 mice; susceptible male x resistant female 89.2 per cent survive for 173 mice. Crosses of resistant males to susceptible females gave progeny which were resistant. This in itself is proof against the transmission of passive humoral immunity from one generation to the next for, as far as is known, males in mammals do not transmit immunity passively to their offspring.

This problem was studied in a different manner by Gowen and Schott (1933c). Susceptible strain females were mated in the same heat period to resistant males and to susceptible males. The two types of progeny in the mixed litters resulting could be separated by their color inheritance; the

FIG. 2. The mouse on the reader's right is from the susceptible strain. It is very sick. The mouse on the left is from a resistant strain. It shows little effect of the disease. Both mice treated alike.

resistant x susceptible progeny were black, those from the susceptible x susceptible parents were silver in coat color. When tested for their typhoid resistance the two types were not identical in their resistance to typhoid, as would be expected if humoral bodies responsible for resistance were passed through to the fetal blood or through the colostrum to the young. The progeny were different in their resistances but different in the manner called for by their inheritance. The F_1's were resistant as was expected. Progeny with only the susceptible inheritance were susceptible.

The results point to cellular differences in the strains as being responsible for the resistance displayed by the strains when in first contact with the typhoid disease.

Tissue Differences of Importance to Typhoid Resistance

The genotypes of these mice evidently play prominent roles in their natural resistances to typhoid. Tissue elements and their immediate reactions are known to be under gene control. The complexity of these reactions is such that it is unlikely that more than a fraction of the pathways by which the gene controls are exerted are uncovered. Attention was called to the leucocytes by Metchnikoff's observation that bacteria may be phagocytosed by them. In the study of the blood of our six strains of mice Gowen and Calhoun (1943) found that particular numbers of both the erythrocytes and the leucocytes were characteristic of each strain. These characters were inherited to the same degree. The erythrocyte numbers of the different strains were distributed at random with regard to the strains' typhoid resistances indicating that they were not of direct importance to the resistance. The leucocytes on the other hand were directly correlated with the resistance. The leucocytes in the resistant strains were more numerous than they were in the strains showing intermediate resistance. In turn, their numbers in the intermediate resistant strains were greater than those in the susceptible strain. Counts of the different kinds of leucocytes were not important. It was as if the leucocytes came from one common stem cell and that it was the capacity of these cells to divide to form large numbers of leucocytes when necessary that was important.

Other fixed cells of the body as the macrophages of the liver and spleen were shown to play a significant role by Oakberg (1946). Another factor appears of importance for these cells. In susceptible mice, bacteria are readily ingested by the macrophages and large numbers of them may be observed within these cells. These bacteria appear normal, have good staining properties and appear to be reproducing normally. In genetically fully susceptible mice the bacteria seem to increase within the macrophages to the point where they may break out of the cell and become new foci of infection. In resistant mice it was difficult to demonstrate macrophages containing ingested bacteria. This was only possible when the dose given was 100 times that received by the susceptible mice. In these cases the bacteria do not stain well and their cell walls appear ragged. It appears as though the macrophages of the resistant lines have a highly effective digestive enzyme which rapidly destroys the ingested *S. typhimuriums*, whereas the enzyme is in reduced amounts or absent in the macrophages of the genetically susceptible mice.

This was not the only difference in cellular reaction that was important, however. The liver cells of the resistant strains will perform the vital functions of the glycogen cycle and in fat synthesis even in the presence of large lesions, whereas the liver cells of the susceptible mice will not. The organ reactions of the resistant are quite different from those of the susceptible mice. Resistant strains may show extensive lesions of the liver and survive, whereas the sus-

ceptible will die without any clinically evident damage to that organ. By contrast, the naturally resistant mice show but little damage to their spleens although the disease may be severe. Susceptible mice, on the other hand, ordinarily display noticeable lesions in this organ. These observations again call attention to the capacities residing within the cells which are of significance to the disease resistance. The liver cells of resistant mice are able to wall off the large necrotic lesion, block off the spread of the *S. typhimurium*, and neutralize any released endotoxins that may be formed thus allowing the remaining tissue to perform its vital functions.

X-ray Irradiation and Typhoid Resistance

The interpretations presented above are subject to further analysis by means of X-ray irradiation of the animal. These observations emphasize the careful consideration that should be given to any X-ray treatment. Gowen and Zelle (1945) X-rayed 789 mice belonging to our six different strains with dosages ranging from 0 to 700 roentgens incident to the body. All six strai reacted in a comparable manner. Radiation decreased the typhoid resistance of the various strains at the same rate per unit of dosage. The numbers of leucocytes of the different strains were decreased to the same relative amounts by comparable X-ray dosages. It is known that total body irradiation will affect many types of body cells. It cannot, therefore, be concluded that the leucocytes are the only cells which have been affected and that they alone are responsible for the changes in resistance. Rather they are an index to what is taking place in cells of like X-ray susceptibility throughout the body. The X-ray treatments reduced the typhoid resistance of the animal and the numbers of its leucocytes proportionally over the full dosage range. While the leucocytes are not the only cells affected these facts support the conclusion that natural disease resistance is dependent on cellular function and on numbers of the phagocytic cells. These relations furnish independent evidence for the conclusions reached by other methods.

Specificity of Genotypes for Natural Resistance

The question arises, is this condition one where the natural resistance extends to one or all diseases, does the individual have an over all constitution or is the constitution specific for each disease? This question was investigated by Gowen and Schott (1933a) for diseases due to *Salmonella typhimurium*, pseudo rabies and the antigenic poison ricin. The genes required for resistance or susceptibility to one disease were independent of those required for resistance to another. Webster (1933) confirmed this observation for the independence of resistance to louping ill and to typhoid. For bacterial species which are related taxonomically the natural resistance to one disease carries over to that due to its close relative. Typhoid resistance is closely correlated with Pasteurella

resistance but less correlated with Klebsiella or pneumococcus resistance. Similar conclusions may be derived from the data of Schutze, Gorer, and Finlayson (1936) for *S. typhimurium*, *S. enteriditis*, louping ill, Pasteurella and pneumococcus.

Taken broadly the results concur in showing that a resistant constitution for one disease is only likely to indicate resistance to another disease if the two diseases are fairly closely related. When the diseases are of different types, resistance to one tends to be independent of resistance to the others. In this respect inheritance for disease resistance behaves like any other inheritance dependent on many genes, some genes are independent, some appear to be linked or to have more than one effect, some have physiological interactions but it would appear that all are ultimately separable.

Genotypes of the Pathogen

The other side of the square portraying the base of the disease syndrome may now be examined. The genotypes of a species of bacteria are as diverse as those of their hosts. They may have more significance to the disease which is produced because they reproduce in such large numbers. The small rate at which mutations of existing genes occur and the rapidity with which such mutants may replace the existing population becomes of great importance in altering the character of the disease produced. The differences may result in changes in the growth pattern of the colony, in the color, morphology, or antigenic properties of the organism. They may affect the cultural requirement of the organism making some strains require a nutrient for growth which other strains can synthesize. The pathogenic characteristics may be altered in either the direction of greater or lesser virulence. The effects of these changes on disease expression have been under study in our laboratory for some 20 years for such diseases as tobacco mosaic, corn wilt, typhoids of both mouse and fowl. The results are concordant.

Changes occur and may readily be established in all of the pathogens involved. The new types may appear at any time during the experiment and may cover a wide range. With selection it is possible to establish many of the new forms as true breeding types. In a given environment competition may exist resulting in rapid replacement of the unfavored types. Changes in virulence as dependent upon the inherited bacterial constitution have been examined by searching out phenotypical variants in originally pure stocks, Zelle (1942), Lincoln and Gowen (1942), Gowen (1948), Plough, Young, and Grimm (1950), and Gowen, Stadler, Plough, and Miller (in Press). The mutant phenotypes are represented by changes in color or morphology of the colonies, antigenic types of the organism, ability to synthesize amino acids or other metabolites from an energy source and simple salts.

The pathogen's ability to initiate a disease in a host is rather highly specific.

As attained in nature this property represents some chance combination of genes which appeared, was preserved, and improved upon by selection during successive generations of reproduction for better and better gene combinations and the inclusion of any of the more favorable mutations which might have occurred as time passed. Contact with an epidemic indicates that an efficient gene combination in the pathogen has evolved. Changes of a random sort comparable to those resulting from chance mutation would be unlikely to improve the disease producing ability of a highly virulent organism. Our results agree with this interpretation. Mutations of our highly virulent lines on the average lead to lines of less virulence. A study of 12 different mutants of 533-11C requiring adenine as a metabolite illustrate this fact. Seven mutants were avirulent causing no deaths, 3 showed some virulence, 2 were as virulent as the parent. The average virulence after mutation was less than the highly virulent parent type. The mutants were separated because of their effects on adenine. The range in virulence shows that this requirement is not itself the primary cause of virulence. Rather it is some change which through chance was associated with the change to adenine requiring that is of importance.

For avirulent cultures any observed changes in virulence will of necessity be toward greater mortality. Mutations of such lowly virulent forms consequently tend to be accompanied by increases in the mortality of the host. The average result of several mutations would be expected to be an increase in virulence. Eleven mutants occurring in our avirulent line 519 illustrate that fact. One of these mutants, that toward leucine requiring, showed a moderate increase in virulence. The rest remained avirulent. The mutations as they are observed are directional in that they tend to be less virulent when their parent is virulent and more virulent when the parent is avirulent. The particular change in observed virulence is not necessarily related directly to the change by which the mutant was detected.

Mutations toward or away from virulence may occur naturally or through the help of radiant energy from X-rays. Quantitative estimates of the frequencies of these changes and of the types of change have been made for several forms. The mutations can likewise affect the capacity of the pathogen to stimulate active immunity in the host.

Acquired Immunity as a Property of Host and Pathogen

Three factors important to increasing resistance to a disease through vaccination are common knowledge. The fourth factor and possibly the most important is not so generally recognized. In order of the emphasis given them, these factors are (1) the dose of the vaccine administered at any one time; (2) the necessity for vaccinations at successive intervals; (3) the line of the bacteria making the vaccine; and (4) the genotype of the host receiving the vaccine.

The first factor takes cognizance of the fact that if the vaccine is too small in amount, it will not stimulate the recipient to form the antibodies of resistance. If on the other hand the dose is too large the endotoxins, etc., released, will be enough to create lesions and sometimes death. The desirable dosage is that quantity which will stimulate a strong immunity without undesirable affects.

The second point is dependent on the fact that as immunization proceeds a greater and greater dose for stimulation can in general be given.

The strain of pathogen is ordinarily considered by the makers of the vaccine. They recognize that some strains of a pathogen are good immunizers and others are not. This is another way of saying that the inheritance of the particular strain of pathogen, its genotype, is of importance to its immunizing properties.

The genotypes of the hosts which are to be immunized up to recently have been neglected.

An experiment, including more than 4,212 mice, was performed to obtain quantitative information on these questions.

Figure 3a shows the relation between immunization dose and the resistance to a dose of 50 million organisms of our highly virulent culture 11C. The chart shows that for this rather large challenge dose less than 50 per cent of the mice are protected. Protection is greater for those mice receiving the most vaccine. In making these immunizations, however, certain mice die presumably due to release of endoxtoxin in the absorbed vaccine. The mice which succumb are those which are least resistant to a first attack of the disease. The genotype of the mouse is correlated with its ability to actively or passively immunize.

Figure 3b shows that three successive immunizations are better than two, and two are better than one. The improvement in resistance with each step is about the same in amount.

Figure 3c presents clear evidence for the significance of the bacterial genotype in immunization. The typhimurium line which displays little virulence was the poorest immunizer. The other two lines having higher virulence were also better immunizers. The avirulent line and one of the virulent lines came from the third line by one step and by two step mutations respectively. As these bacterial lines come from each other by mutation, the importance of even a gene difference in the lines' genotypes is evident.

Figure 3d shows the effects of the hosts' genotypes on their abilities to immunize. The six different strains immunize differently. The naturally resistant strains on first contact with the disease are those which have their resistances enhanced most by immunization. The intermediate strains, as measured by natural resistance, are likewise intermediate in their abilities to immunize. The most susceptible strains after immunization remain more susceptible than the other genotypes immunized in like manner. The level of resistance of each strain is simply raised a proportionate amount. In terms of

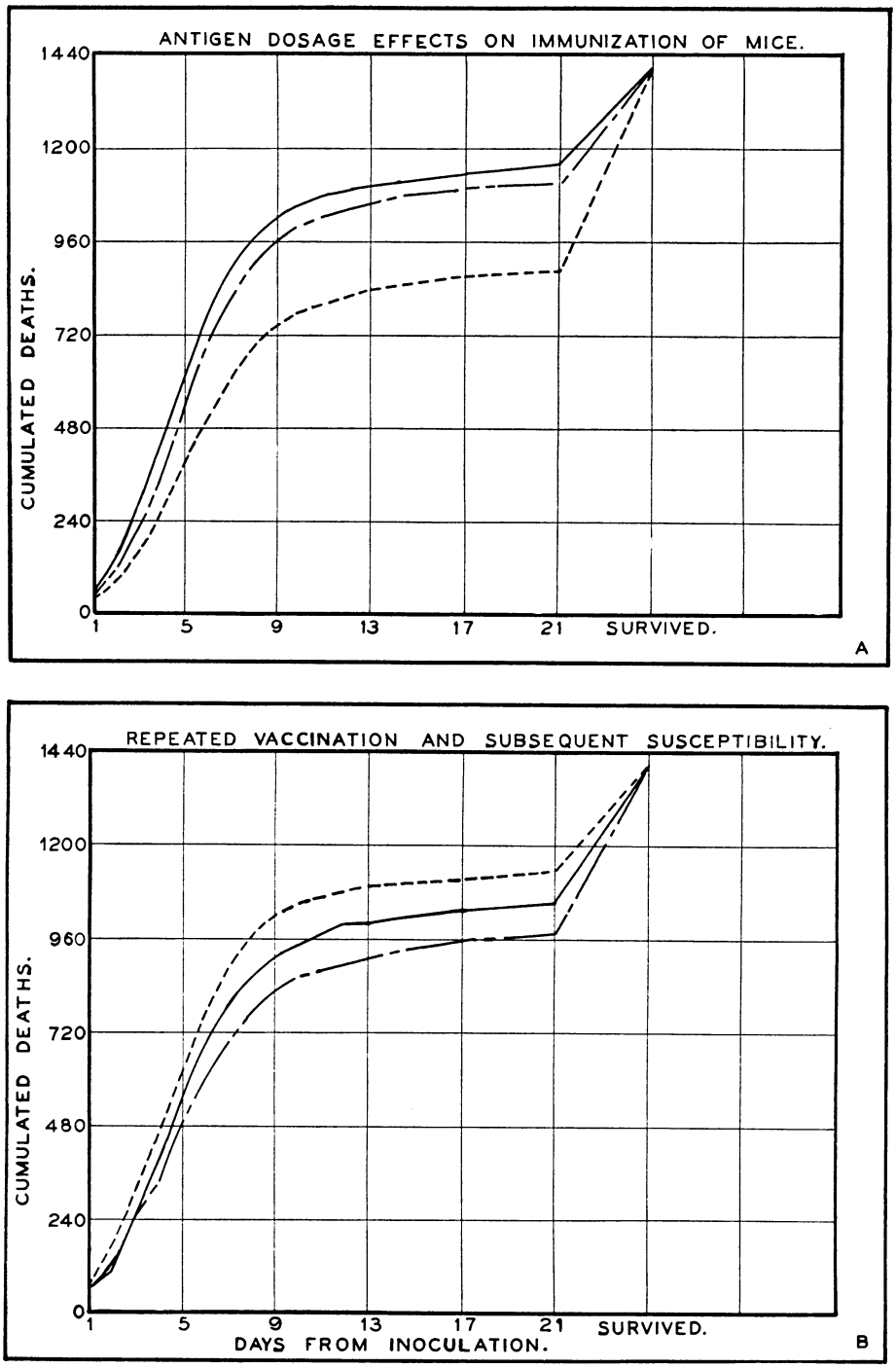
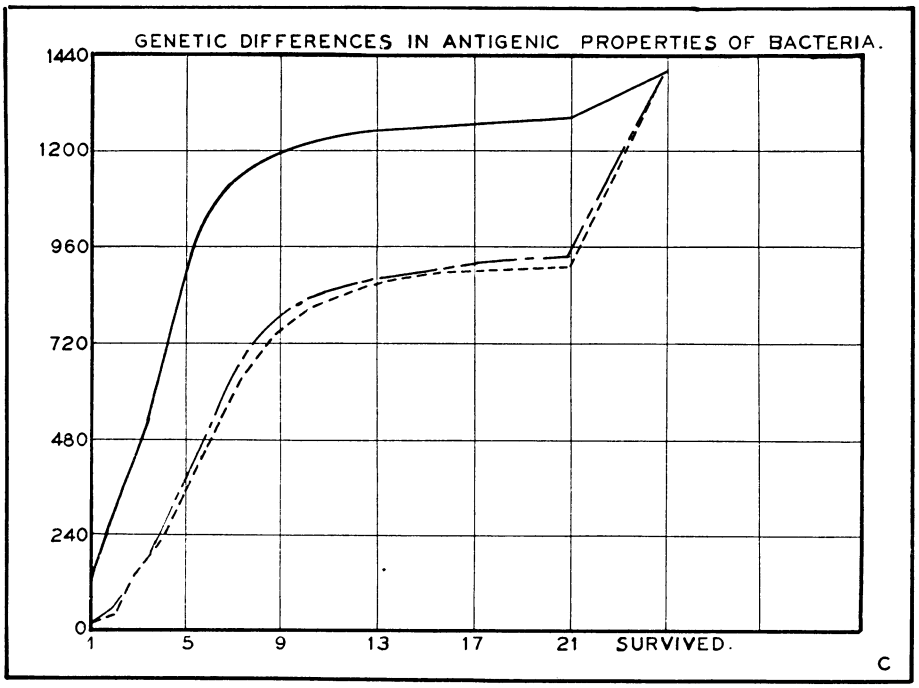


FIG. 3. Resistance acquired through vaccination with heat killed cultures of *Salmonella typhimurium* 11C as related to (a) amount of vaccine administered; (b) times vaccinated; (c) genotypes of bacteria furnishing the vaccine; (d) host's genotype for natural resistance. Graphs show cumulated deaths from inoculation by days during course of disease.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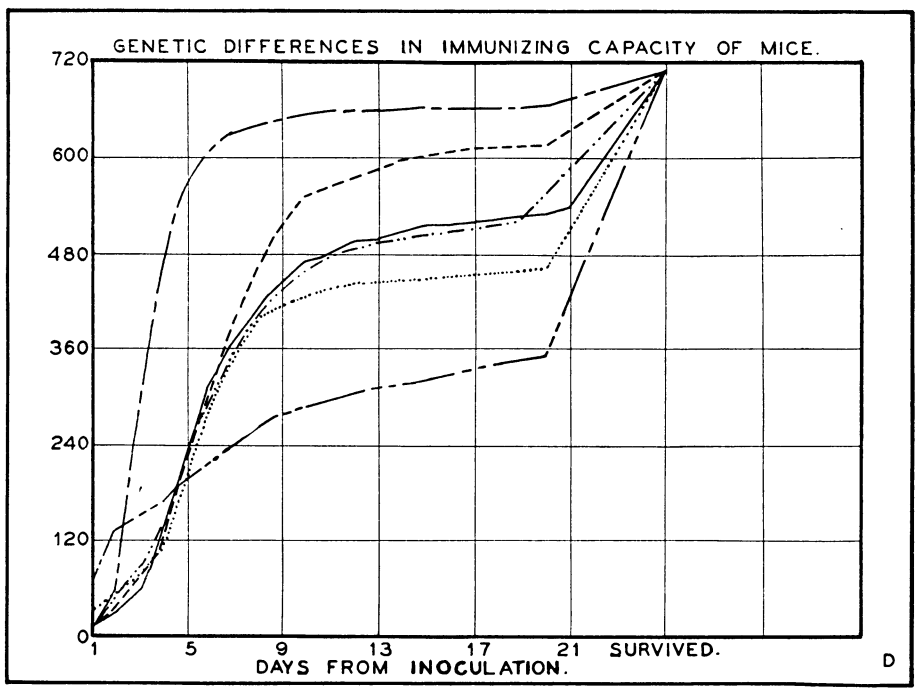
GENETIC DIFFERENCES IN IMMUNIZING CAPACITY OF MICE.
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FIG. 3

bacteria which may be inoculated, immunized mice can resist 100 to 200 times as many live virulent strain bacteria as the unvaccinated mice.

In our mice cellular elements show a high correlation with natural resistance. Humoral elements are not found prior to the first contact with the disease. They are not part of the phenotypes of the different strains but are only called out by contact with the pathogens. The effects of the vaccinations parallel those found for natural resistance in that they depend on the genotypes of the hosts and pathogens. These facts suggest that the results attained in acquired immunity to typhoid are due to attributes of the cellular mechanisms which are likewise responsible for natural resistance.

APPLICATIONS TO DISEASE CONTROL

By successive steps medical science has advanced from a concept of disease as due to a single demon as Apollo's arrows, having as its purpose the punishment of man, through the Hippocratic differentiation of disease into different and distinct complexes based upon their clinical symptoms, to the concept that diseases are truly separable and open to proper treatment when the initiating and perpetuating agent or agents are known. Genetical research is carrying forward the traditional objectives of this unfolding thought. Through efforts of workers in the field it is now recognized that agents of diseases and their hosts each are organized and get their characteristics from the development of an inheritance, made up of distinct genes. By specifying the gene for a particular disease reaction it has been possible to show that genes act as protectors from a given disease, as independent of the disease reaction, and as causative agents in the disease syndrome. A clinically manifest disease only results when the proper combination of the genotype of the victim, the genotype of the pathogen, where one is necessary, are properly synchronized with the environment.

This synchronization is illustrated earlier by the full and no complement genes in guinea pigs. The full complement gene governs the formation of normal complement, a humoral substance having several important disease protecting properties. The no complement allele forms a substance like complement in all but one component. That component is essential for resistance to two of the guinea pig's pathogens as well as to the binding of antibody and antigen. These genes operate through a change in a humoral body which they have a part in forming.

The localization of the genes has shown, as with diseases in which pathogens are responsible, that clinically identical symptoms may be due to different causes operating through distinctly different channels. This fact issues its own warning for as Haldane (1948) says "just as the methods for the cure of bacillary and amoebic dysentery are very different, so it is unlikely that the same therapeutic measures will succeed against diseases, however similar in their symptoms, which are due to different genes." Considerations of this kind

emphasize the need for careful study of all cases where inheritance plays a major role in the initiation of the disease.

In considering the probable severity of a disease attack something is often known on the incidence and the severity with which that disease has appeared previously in the family. This information may have considerable value in prognosis. The disease may be detected at an earlier stage than otherwise, making treatment more effective. Information on the inheritance patterns in our mice not only give information on the mortality but it also indicates the severity of the reactions at each successive stage in the disease cycle. Physical characters like weight, muscular activity normally considered in estimation of physical constitution, have little bearing on the outcome. Normal body temperature levels of the mouse do not appear to influence the course of the disease, although in fowl typhoid a fraction of a degree difference in normal temperature appears important. The characteristics of the normal blood have prognostic value. In normal mouse and chicken blood, humoral elements related to their typhoid resistance have been found lacking. The leucocyte number for our mice is related to the observed resistance but is not the whole story, for the phagocytic cells of different strains differ in their ability to destroy the typhoid organism.

The results have a direct bearing on the explosive appearance of a disease in a population (Gowen 1951). The factor making toward this sudden appearance appears to be mutation toward greater virulence of the pathogen. This has been demonstrated several times in *Salmonella typhimurium*, *Salmonella gallinarum*, and in *Phytomonas stewartii*, where saprophytic lines have suddenly changed to virulent forms. The reverse change of virulent to saprophytic types is also demonstrated. Such changes would form a means of preserving the organisms over unfavorable times. The influence of these factors has had unwitting demonstration under field conditions in our major food crops. The factors involved are a host of uniform genotype, contact between individuals sufficiently close to make for easy spread of a particular disease, a pathogen whose genotype fits that of the host in a manner favorable to invasion, high morbidity and mortality. These conditions have been satisfied repeatedly particularly in our cereal crops. In fact, during the last fifty years the conditions have been fulfilled by a succession of quite diverse pathogens able to invade in succession the newer synthesized varieties built up to resist older diseases. In succession strains of rusts and smuts, *Helminthosporium victoriae* and a previously little known crown rust (Murphy 1948–49) have assumed the major pathogen role.

Each host shows a cyclic rise of infection and mortality. Each cycle is highly dependent upon how well the genetic constitutions of the particular host and pathogen fit together. The necessary close contact between susceptible individuals is established by the uniform planting of the same host genotype

over large areas of the suitable agricultural lands. Dosage with the virulent disease is high because of the close contact. Because of the close association of individuals, the susceptible host genotypes, the virulent genotypes of the pathogens, and the large amount of inoculum received by each individual, epidemics of major severity follow.

Considered individually each epidemic comes near meeting the simplest postulates yet devised to account for the rise in epidemic mortality, Brownlee (1906–10), Ross (1915), Soper (1929), Frost (1941), and Wilson (1945). In succession, each different population is composed of a single genotype whose individuals will have identical susceptibilities for a pathogen of a particular genotype. In each case a very small number of the pathogens come into or exist in the region. Because of the host genotype on which to work and the dense plantings of this favorable host making for easy spread, the rare pathogen is able to rapidly outgrow all other types and create massive infections of any remaining plants. The resulting mortality decimates the host. By man's efforts the first host is replaced by a host of genotype resistant to the first disease but susceptible to another which is present in the region again in small numbers. The process is repeated. The successive changes of the host genotypes create successive cycles, first favorable to the host's reproduction and survival, then favorable to the particular pathogen and high host mortality.

In animal or human populations where the genotypes are diverse the above conditions are only partially fulfilled and the severity of the epidemic is consequently modified. The unconscious approach to the ideal condition as found in our cereal crops helps to confirm the significance of host and pathogen genotypes in disease expression.

SUMMARY

This paper discusses the part played by the genes, making up the genotypes of both host and pathogen, in disease causation and severity. Gene differences within a host may act as pathogens to their host, in the sense that they may cause recognizable syndromes, and may spread through successive generations of the population. Gene differences between different members of the host population may be responsible for susceptibility to a given pathogen on the one hand and resistance to the same pathogen on the other. Such differences may be due to a single pair of alleles as for Pasteurella of the guinea pig or to many pairs of genes as in typhoid of the mouse. Genes affect different features of the natural resistance pattern. The paths through which the effects are produced are very imperfectly surmised. Specificity is ordinarily observed for the gene effects on resistance particularly if the pathogens initiating the diseases are taxonomically rather far removed from each other. The specificities of the gene effects for a given disease under natural conditions have most

frequently been traced to cellular differences with humoral differences occurring as a secondary consequence of cellular activity.

Genotypic differences are displayed by individuals within a pathogenic species. These differences modify virulence. Like differences in genotype affect the host's property of acquiring immunity on exposure to a disease. The interactions of the host genotypes with those of the pathogen are basic not only to the initial severity of the disease but also to the level of immunity acquired after the survival of the initial attack. In disease, as it occurs naturally, full expression of these interacting factors together with those characteristic of the environment ordinarily lead to a great variety and complexity of disease reactions. Under certain conditions primary individual factors may be simplified, for example, in an epidemic of cereals, to the point where the parts of the complex interacting system of the disease processes may be recognized and evaluated in terms of each contributing element. When so observed epidemics appear to be generated, to rise in severity, and to fall, as the result of a dense population of susceptible genotypes having in it a few pathogens of the genotype for high infectivity and ordinarily high pathogenicity and virulence for the host susceptible genotype in an environment which leads to selective rapid reproduction of the virulent pathogenic strain and to massive doses to the host population.

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3. On the Mechanism of Genetic Resistance to Tuberculosis and its Mode of Inheritance[1,2]

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OVER the past 20 years, by bother and sister inbreeding of rabbit stocks for over eight generations and subsequent intrafamilial propagation, various races have been developed. These exhibit different native heritable resistance

[1] Paper presented as part of Symposium: "Light from Animal Experimentation on Human Heredity" at the fifth annual meeting of The American Society of Human Genetics, Ithaca New York, September 9, 1952.

[2] Aided by grants from the Commonwealth Fund, The National Tuberculosis Association and The Public Health Service, Federal Security Agency.

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